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**ABSTRACT** Analysis of morphometrics and allozyme variation were performed to estimate the genetic and morphological variation and to assess both the phylogenetic relationships and biogeographic distribution of the *Pheidole fervida* in Korea. The relationships among populations of this species, in six regions, were also investigated based on the data collected from Mar. 1993 to Oct. 1994. In addition, quantitative morphological data were analyzed, as were genetic variances of allozyme based on electrophoretic data by SGE. As a result of comparing both the morphological and electrophoretic phenograms, it was found that Chilgapsan and Chiaksan populations were genetically closer ( $D = 0.335$ ) than those between others. Naksansa and Yeosu populations were genetically remote ( $D = 1.167$ ) from each conspecific population.

**Key words :** Ants, *Pheidole fervida*, Genetic Variation, Electrophoretic, SGE

Since the original demonstration of extensive genetic variation of enzyme-encoding loci in natural population (Lewontin and Hubby, 1966), one of the primary goals of population genetic has been to identify the forces responsible for this variation (Lewontin, 1974; Nei, 1975; Ayala, 1976; Nei and Koehn, 1983). In the past decade, an increasing number of studies have attempted to identify directly biochemical differences among allozymes as part of multilevel investigation of the adaptive basis of allozyme variation (Koehn et al., 1983).

Genus *Pheidole*, of which four species have been recorded in Korea, is a world wide genus with a large number of species belonging to the subfamily Myrmicinae. In Korea, the genus has been investigated by some researchers, Teranishi (1940), Kim (1963, 1970), Kim (1982), Terayama et al. (1992), Choi (1993), and Kim et al. (1993, 1994). Since populations of this species vary in morphology and colour, we carried out the analysis of intraspecific variation by quantitative analysis of morphological data and the analysis of isozyme variation by SGE (Selander et al., 1971).

### MATERIALS AND METHODS

All the specimens used in this study were from

six localities in Korea. Whenever ants' nests were found in the fields or mountainsides, they were opened with a spade. Some of the collected specimens were put into Bouin's alcoholic solution (150 ml of 80% ethanol, 15 ml formaldehyde and 1 g picric acid), and transferred 2~3 days later to 95% ethanol. Others, intended for electrophoretic use, were placed on liquid nitrogen tank or ice box, and later moved to the deep freezer at  $-60^{\circ}\text{C}$ .

Dried specimens were microscopically examined using the stereo zoom-microscope (Wild M8), and identified in comparison with voucher specimens from Collingwood in Great Britain, Onoyama in Japan, Rigato in Italy and Wang in China, as well as the type specimens from the City Museum Leeds, U.K. Once identified, the specimens used in this analysis were soldier caste because worker was too small to analyze for morphometrics. A total of 20 individuals per regions were measured under the stereo zoom-microscope for head width (HW), head length (HL), eye width (EW), eye length (EL), scape length (SL), cephalic index ( $\text{CI} = \text{HW}/\text{HL} \times 100$ ), and scape index ( $\text{SI}① = \text{SL}/\text{HL}$ ;  $\text{SI}② = \text{SL}/\text{HW}$ ). The coefficients of similarity between six regions were offered using unweighted pair group method of arithmetic average (UPGMA) by SPSS/PC+, and following the Elmes (1978). For electrophoresis of allozyme variation, approximately hundred or more individuals per population were

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used, considering individual variation regarding polymorphism. After performing starch gel electrophoresis (SGE; Selander, 1971), gels were obtained. The degrees of genetic variation based on the estimated NTSYS program, used UPGMA, WPGMA, COMPL.

## RESULTS

### Description

*Pheidole* Westwood, 1840, 6: 87.

Type species : *Atta providens* Sykes, 1835.

### *Pheidole fervida* Fr. Smith, 1874 극동흑개미

*Pheidole fervida* Fr. Smith, 1874: 406; Forel, 1900: 285; Teranishi, 1940: 4; Ogata, 1982: 195; Imai, 1966: 119.

*Pheidole fervida* var. *kwazana* Teranishi, 1940; ?; Kupyanskaya, 1990: 124.

*Pheidole fervida*; Kim, 1963: 344; Kim, 1970: 477; Collingwood, 1976: 302; Kim and Kim, 1983: 71; Terayama et al., 1992: 27; Kim et al., 1993: 124; Kim et al., 1994: 299; Kim et al., 1995: 105; Kim et al., 1996: 122.

### Diagnostics Description; Dimorphic species.

Large worker (Soldier); Body length 3.7 mm to 4.7 mm. Head, thorax and petiole brownish yellow, abdomen brown, legs and antennae yellow, mandibles dark brown. Head length more or less longer than width, and a large number of furrows distinct toward the frontal margin. Occipital margin subsided deeply, carina indistincts. Clypeus plate and anterior margin concaved deeply. Compound eyes small and located forward. Mandibles two apical teeth widely from small basal tooth. Antennae 12 segmented with 3 apical segments enlarged to distinct club. Pronotum rounded anterolaterally, width two times as head width in dorsal view. Connected area between metanotum and mesonotum subside but indistinct. Spine on propodeum short, back-upward in lateral view. Petiole peaked or rounded in profile. Postpetiole without distinct ventral projection, width two times as petiole width. Both thorax and petiole indistincts furrows irregularly. Abdomen egg-shaped.

Small worker (Worker); Body length 1.3 mm to 1.58 mm. Head, thorax and petiole brownish yellow, mandibles, antennae and legs light brown, abdomen dark brown. Long oval heads multidentate

mandibles. Mandibles two apical teeth widely from small basal tooth. Antennae 12 segmented with 3 apical segments enlarged to distinct club. Antennal scape much extending backward beyond posterior area of head. Pronotum width narrower than occipital width. Others part resembled soldiers.

**Measured Value.** EL = 0.313–0.338, EW = 0.205–0.229, HL = 2.471–2.653, HW = 2.403–2.557, SL = 1.295–1.391, CI = 97.228–99.033, SI① = 50.715–54.510, SI② = 51.925–55.059.

**Specimens Examined.** Deokyusan (3.X.1994), Jirisan (22.VIII.1994), Yeosu (29.IV.1994), Naksansa (15.VI.1994), Chiaksan (20.V.1994), Chilgapsan (29.VII.1993).

**Localities.** Gaeseong, Bakyon-Pokpo, Sagampo, Geumgangsan, Pyeongyang, Seokwangsa, Ganghwado, Seoraksan, Seoul, Songnisan, Woraksan, Gyeryongsan, Jirisan, Namhaedo, Geojedo, Naejangsan, Odongdo, Yokjido, Hansnado, Hanllasan.

**References Record.** Terayama (1940), Collingwood (1976).

**Distribution.** Korea, Japan, China.

### Comparison between morphological and electrophoretic analysis

The result of morphological analysis, which depends on degree of similarity based on the external morphological characters showed that they were significantly different among them. The degree of similarity between Deokyusan and Jirisan (similarity = 95), Chiaksan and Chilgapsan (similarity = 90) were morphologically higher than those between others, between Naksansa and others were lower (Table 1, Fig. 1).

On the other hand, the results of electrophoretic analysis, using SGE are a little different from morphological characters but similar to morphological analysis. The degree of genetic variation of Naksansa ( $P = 0.3818$ ,  $H_0 = 0.0317$ ,  $H_E = 0.1845$ ) was lower than those of others. Deokyusan ( $P = 0.5934$ ,  $H_0 = 0.0814$ ,  $H_E = 0.2769$ ) showed highest variability (Table 2–5; Figs. 2, 3).

Based on the estimated NTSYS program, used UPGMA, WPGMA and COMPL, it seemed that Chilgapsan and Chiaksan population were genetically closer ( $D = 0.335$ ) than those between others, Naksansa and Yeosu population were genetically remote ( $D = 1.167$ ) from each conspecific population.

**Table 1.** Morphological characters measured of *Pheidole fervida* (Soldier). Values are given as modes and in parentheses, minima, maxima. (Populations: 1. Naksansa; 2. Chiaksan; 3. Chilgapsan; 4. Deokyusan; 5. Jirisan; 6. Yeosu)

	EL	EW	HL	HW	SL	CI	SI①	SI②
1	0.328 (0.31~0.35)	0.221 (0.20~0.24)	2.653 (2.47~2.62)	2.513 (2.45~2.60)	1.391 (1.37~1.41)	99.033 (97.31~102.07)	54.510 (53.05~55.92)	55.059 (53.46~56.32)
2	0.338 (0.32~0.36)	0.224 (0.20~0.25)	2.565 (2.40~2.70)	2.557 (2.43~2.65)	1.373 (1.32~1.40)	97.736 (96.15~108.33)	53.569 (50.00~57.50)	53.755 (51.53~55.87)
3	0.329 (0.30~0.35)	0.216 (0.20~0.23)	2.589 (2.30~2.70)	2.548 (2.40~2.70)	1.352 (1.29~1.40)	98.427 (93.23~108.00)	52.140 (47.78~56.33)	53.323 (48.31~57.83)
4	0.313 (0.30~0.35)	0.215 (0.20~0.35)	2.515 (2.34~2.83)	2.459 (2.31~2.80)	1.317 (1.28~1.34)	97.751 (94.62~99.20)	52.444 (46.64~54.58)	53.662 (47.14~56.71)
5	0.319 (0.30~0.34)	0.205 (2.20~0.22)	2.471 (2.30~2.62)	2.403 (2.30~2.56)	1.307 (1.27~1.36)	97.228 (95.00~100.00)	52.980 (50.00~56.52)	54.346 (50.98~56.70)
6	0.330 (0.30~0.38)	0.229 (0.21~0.25)	2.555 (2.40~2.70)	2.495 (2.31~2.60)	1.295 (1.28~1.32)	97.729 (95.35~100.81)	50.715 (48.52~53.33)	51.925 (49.61~55.41)

**Table 2.** Allele frequencies of polymorphic loci of *Pheidole fervida*. Numbers refer to the collection regions listed in Table 1. (S : soldier, W : worker)

Locus	allele	1		2		3		4		5		6	
		S	W	S	W	S	W	S	W	S	W	S	W
Mdh-1	a		.30	.25	.25	.25	.25	.25	.25	.25	.25	.25	.25
	b		.30	.05	.05	.05	.05	.05	.05	.05	.05	.05	.05
	c	.40											
	d	.60	.40	.70	.70	.70	.70	.70	.70	.70	.70	.70	.70
Mdh-2	a		1.00				1.00	1.00					
	b	1.00		1.00	1.00	1.00			1.00	1.00	1.00	1.00	1.00
Me	a	1.00	1.00				1.00						
	b			1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00
Idh	a								1.00			1.00	
	b			1.00	1.00	1.00	1.00	1.00		1.00	1.00		1.00
Akp-1	a							.30	.30				
	b	1.00	1.00	1.00	1.00	1.00	1.00	.06	.10				
	c							.64	.60	1.00	1.00		
Akp-2	a		1.00		1.00		1.00		1.00				1.00
Acp	a			.50	.50	.50	.50						
	b			.50	.50	.50	.50	.50	.50				
	c	1.00	1.00					.50	.50				
Pt-1	a	.50	.50	.50	.50								
	b					.49	.49	.45	.45				
	c									.40	.40	.42	.42
	d			.10	.10	.02	.02	.06	.06	.10	.10	.10	.10
	e	.50	.50	.40	.40								
	f					.49	.49	.49	.49				
Pt-2	a			.25						.50	.50	.48	.48
	b				.36								
	c			.05		.25	.24	.16	.16				
	d									.36	.20	.25	.25
	e					.11	.06	.24	.24	.05			
	f			.70	.64	.64	.70	.70	.70	.64	.75	.75	.75
Got-1	a	1.00	1.00										
	b			1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00
Got-2	a	.20	.20	.15	.15	.15	.15	.15	.15	.15	.10	1.00	1.00
	b	.80	.80	.85	.85	.85	.85	.85	.85	.85	.90		
Est-1	a			.50	.50								
	b	1.00	1.00			1.00	1.00	.40	.50		1.00		
	c			.50	.50			.60	.50				
Est-2	a	.40	.40	.36	.36	.40	.40						
	b							.45	.45	.40	.40	.36	.36
	c	.10	.10	.14	.14	.10	.10						
	d							.05	.05	.10	.10	.14	.14
	e	.50	.50	.50	.50	.50	.50						
Est-4	f							.50	.50	.50	.50	.50	.50
	a			1.00	1.00	1.00	1.00	1.00	1.00			1.00	1.00
	b									1.00	1.00		

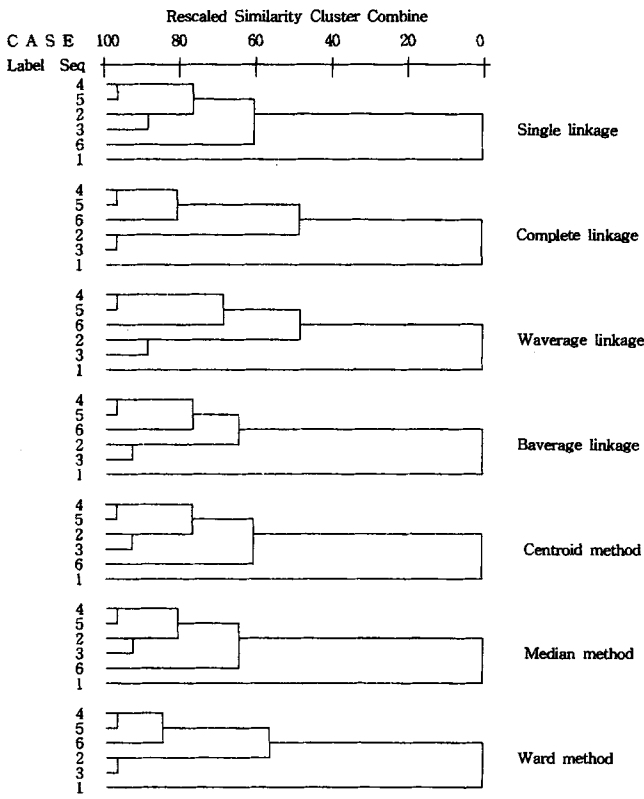


Fig. 1. Phylogenetic relationships of 6 populations in *Pheidole fervida* (Soldier) generated by analysis of morphological characters.

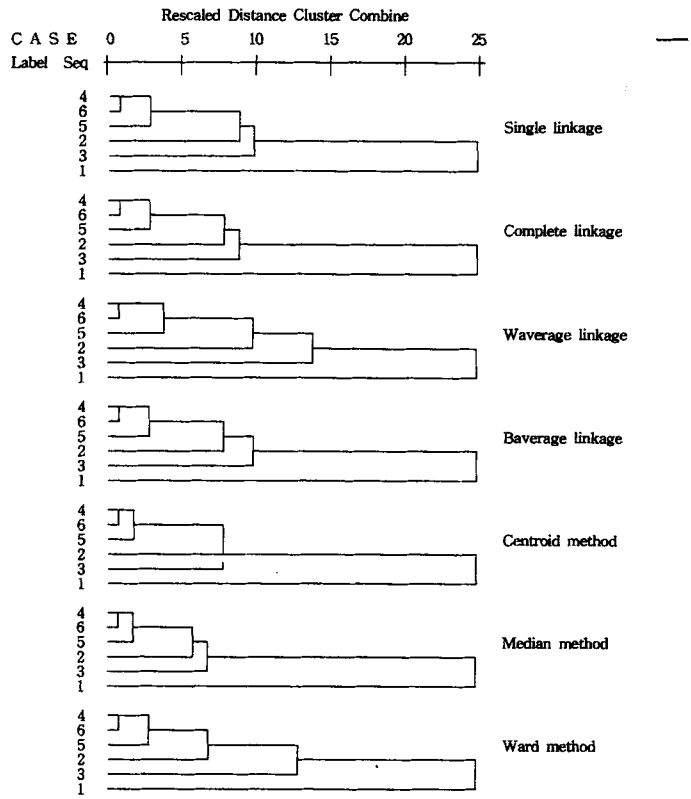


Fig. 2. Phylogenetic relationships of 6 populations in *Pheidole fervida* (Soldier) generated by analysis of electrophoretics.

Table 3. Nei's (1972) genetic dissimilarity coefficients of *Pheidole fervida*

A

	1	2	3	4	5	6
1 Naksansa		0.905	0.434	1.097	1.483	1.727
2 Chiaksan	0.799		0.358	0.351	0.563	0.309
3 Chilgapsan	0.693	0.135		0.556	0.770	0.657
4 Deokyusan	1.425	0.391	0.331		0.447	0.367
5 Jirisan	1.372	0.465	0.498	0.433		0.436
6 Yeosu	1.604	0.518	0.554	0.616	0.491	

B

	1	2	3	4	5	6	7	8	9	10	11	12
1 Naksansa-S												
2 Naksansa-W	0.224											
3 Chiaksan-S	0.799	1.148										
4 Chiaksan-W	0.847	0.905	0.060									
5 Chilgapsan-S	0.693	0.999	0.135	0.189								
6 Chilgapsan-W	0.742	0.434	0.441	0.358	0.265							
7 Deokyusan-S	1.425	1.094	0.391	0.447	0.331	0.377						
8 Deokyusan-W	1.038	1.097	0.441	0.351	0.359	0.556	0.296					
9 Jirisan-S	1.372	2.079	0.466	0.519	0.498	0.969	0.435	0.487				
10 Jirisan-W	1.039	1.483	0.507	0.563	0.382	0.770	0.408	0.447	0.056			
11 Yeosu-S	1.604	2.659	0.518	0.575	0.554	1.077	0.616	0.307	0.491	0.554		
12 Yeosu-W	1.659	1.727	0.404	0.309	0.438	0.657	0.483	0.372	0.372	0.436	0.194	

A; among the 6 regions Soldier (below diagonal) and Worker (above diagonal)  
B; among the 12 populations

**Table 4.** Observed heterozygosity ( $H_O$ ) and expected heterozygosity ( $H_E$ ) at 14 loci of *Pheidole fervida*

Locus	Naksansa				Chiaksan				Chilgapsan			
	Soldier		Worker		Soldier		Worker		Soldier		Worker	
	$H_O$	$H_E$	$H_O$	$H_E$	$H_O$	$H_E$	$H_O$	$H_E$	$H_O$	$H_E$	$H_O$	$H_E$
Mdh-1	0.0000	0.4800	0.1723	0.5900	0.0515	0.4450	0.0515	0.4450	0.0515	0.4450	0.0515	0.4451
Mdh-2	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000
Me	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000
Idh			0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000
Akp-1	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000		0.0000	0.0000
Akp-2			0.0000	0.0000			0.0000	0.0000			0.0000	0.0000
Acp	0.0000	0.0000	0.0000	0.0000	0.0000	0.5000	0.0000	0.5000	0.0000	0.5000	0.0000	0.0000
Pt-1	0.0000	0.5000	0.0000	0.5000	0.2226	0.5800	0.2226	0.5800	0.1765	0.5194	0.1765	0.5194
Pt-2					0.2510	0.4450	0.2510	0.4450	0.3000	0.5158	0.3000	0.4488
Got-1	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000
Got-2	0.0000	0.3200	0.0000	0.3200	0.0000	0.2550	0.0000	0.2550	0.0000	0.2550	0.0000	0.2550
Est-1	0.0000	0.0000	0.0000	0.0000	0.0000	0.5000	0.0000	0.5000	0.0000	0.5000	0.0000	0.5000
Est-2	0.2500	0.5800	0.2500	0.5800	0.3500	0.6008	0.3500	0.6008	0.2533	0.5800	0.2533	0.5800
Est-4					0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000
Aver.	0.0250	0.1880	0.0384	0.1809	0.0673	0.2558	0.0466	0.2387	0.0673	0.2166	0.0648	0.1963

Locus	Deokyusan				Jirisan				Yeosu			
	Soldier		Worker		Soldier		Worker		Soldier		Worker	
	$H_O$	$H_E$	$H_O$	$H_E$	$H_O$	$H_E$	$H_O$	$H_E$	$H_O$	$H_E$	$H_O$	$H_E$
Mdh-1	0.0000	0.4800	0.1723	0.5900	0.0515	0.4450	0.0515	0.4450	0.0515	0.4450	0.0515	0.4451
Mdh-2	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000
Me	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000
Idh	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000
Akp-1	0.0333	0.4968	0.2510	0.5400	0.0000	0.0000	0.0000	0.0000	0.0000		0.0000	0.0000
Akp-2			0.0000	0.0000							0.0000	0.0000
Acp	0.0000	0.5000	0.0000	0.0000							0.5000	0.5000
Pt-1	0.3000	0.5538	0.3000	0.5538	0.3333	0.5800	0.3333	0.5800	0.3621	0.5832	0.3621	0.5832
Pt-2	0.3511	0.4268	0.3511	0.4268	0.0000	0.4608	0.0025	0.3950	0.0000	0.3750	0.0000	0.3750
Got-1	0.0000	0.0000	0.0000	0.0000	0.0300	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000
Got-2	0.0000	0.2550	0.0000	0.2550	0.0000	0.2550	0.0000	0.1000	0.0000	0.0000	0.0000	0.2550
Est-1	0.0000	0.4800	0.0000	0.5000			0.0000	0.0000				
Est-2	0.2632	0.5450	0.2632	0.5450	0.2531	0.5800	0.2531	0.5800	0.3520	0.6008	0.3520	0.6008
Est-4					0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000
Aver.	0.0769	0.2848	0.0869	0.2690	0.0930	0.2110	0.0852	0.1750	0.0765	0.2004	0.0696	0.1822

\*  $H_E$ ; calculated on the basis of the allele frequencies.

$$H = 1 - \sum X_i^2, \text{ where } X_i \text{ is frequency of the } i\text{th allele at locus.}$$

\*  $H_O$ ; fraction of heterozygotes individuals.**Table 5.** Genetic variation of the *Pheidole fervida*

	Mean No. of allele per locus (A)	% Polymorphism per locus (% P)	Heterozygosity	
			Observed ( $H_O$ )	Expected ( $H_E$ )
1 Naksansa-S	1.50	40.00	0.0250	0.1880
2 Naksansa-W	1.55	36.36	0.0384	0.1809
3 Chiaksan-S	1.85	53.85	0.0673	0.2558
4 Chiaksan-W	1.71	50.00	0.0446	0.2387
5 Chilgapsan-S	1.77	46.15	0.0673	0.2166
6 Chilgapsan-W	1.71	42.86	0.0648	0.1963
7 Deokyusan-S	2.00	61.54	0.0769	0.2848
8 Deokyusan-W	1.93	57.14	0.0869	0.2690
9 Jirisan-S	1.73	45.45	0.0930	0.2110
10 Jirisan-W	1.75	41.67	0.0852	0.1750
11 Yeosu-S	1.70	40.00	0.0766	0.2004
12 Yeosu-W	1.64	36.36	0.0696	0.1822
	1.74	45.95	0.0663	0.2166

## DISCUSSION

Samples of a species taken from different areas may differ significantly in their allelic frequencies. This could be due to selection for different homozygotes under varying environmental conditions or to genetic drift in isolated populations. In a wide spread panmictic population, selection may produce differential survival in a different regions and result in allelic frequency variation. Since allozyme have different structures they may be detected electrophoretically as bands of different mobility which react with the same substrate. From the systematist's point of view, the existence of isozymes means that care must be taken to examine hom-

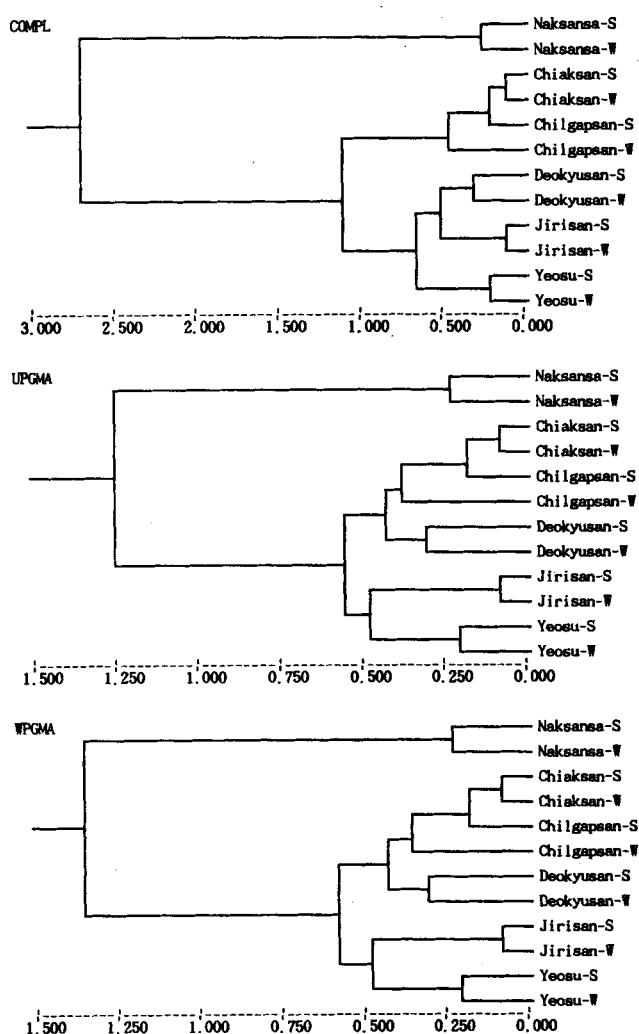


Fig. 3. Dendrograms of *Pheidole fervida* based on Nei's genetic dissimilarity treated with NTSYS (S; soldier, W: worker)

ologous life stages, tissues and parts of cells when making inter-taxon comparisons (Ferguson, 1980).

Since there are environmental and nongenetic influences, the systematics dependant on only morphological characters could cause a lot of problems obscuring their phylogenetic relationships among members of the group. Thereupon, it is generally accepted that allozyme variation from electrophoretic information is resonable for analysis of intra-specific variation.

Comparing the phenograms of morphological variation with those of allozyme variation, it seemed that two results agreed with each other nearly, while the degree of similarity between the group Naksan-sa and others were quite different. We conclude that the six populations of *Pheidole fervida* in Korea have following relationships; Chilgap-

san and Chiaksan populations have 94% degree of similarity; Deokyusan and Jirisan populations have 88%; they and Yeosu populations have 80%. Naksansa population were the farthest population from others in morphological and genetical tracts.

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